REMARKS

Claims 1, 17, 19-24, 45, 57-59, 61, 62, 67, and 77 are pending.

Claims 17 and 19 have been cancelled herein without prejudice.

Specification objections

Applicants have amended the specification as described above in view of the Examiner's comments.

Applicants, therefore, respectfully request withdrawal of the specification rejections.

Rejections under 35 U.S.C. § 112, second paragraph

The Examiner has rejected claims 20, 45, 57-59, 61, 62, 67, and 77, under 35 U.S.C. § 112, second paragraph, as allegedly being indefinite for reasons as stated in the Office Action.

Applicants have responsively amended the claims to obviate these rejections, and therefore respectfully request withdrawal of these rejections.

Rejections under 35 U.S.C. § 112, first paragraph

The Examiner has rejected claims 1, 17, 20-24. 45, 57-59, 61, 62, 67, and 77, under 35 U.S.C. § 112, first paragraph, as allegedly lacking sufficient enablement.

Specifically, the Examiner, citing the *Wands* factors (claim scope; nature of invention; state of the prior art; level of skill in the art; level of predictability; amount of direction provided; working examples; and quantity of experimentation required), states with respect to claim scope that Applicants' claims broadly encompass methods to predict responsiveness of *any* subject, to a wide variety of drugs that differ in structure and mechanism of action, in which the drug is used to treat *any* breast tissue proliferative disorder (broad genus as recited in the Office Action) that may differ with respect to symptoms and etiology, and additionally include analysis of *any* biological sample (e.g., frozen tissue, paraffin embedded from any organ, acellular samples, etc.) for CpG methylation of *any* sequence of the PITX2 gene, and finally broadly recite determining

methylation status wherein predicting responsiveness of the subject to the therapy is afforded, without setting forth how the results of "determining" are used for such prediction.

The Examiner states that the invention is that of an "unpredictable" class. The Examiner states that "criteria for defining responders versus non-responders is unclear." The Examiner states the figures do not identify the particular nucleotide position of the CpGs within the PITX2 gene or the amplified fragment of the PITX2 gene." The Examiner states that subjects (Data set 4) "are not clearly characterized with respect to their disorder or as to whether N is the number of each of the responder and non-responder subjects or N is the combined number of responder." The Examiner states that PITX2 is not included in a list of 5 "independent predicting genes" listed on page 67. The Examiner accordingly states that the specification is not sufficiently complete or detailed to allow one to interpret the information (e.g., sample source, particular CpGs, increase or decrease of methylation).

The Examiner states that post-filing art of Martens et al. (Cancer Research, 65:4104-41-7, 2005), and Nimmrich et al (Breast Cancer Research and Treatment, 111:429-437, 2008) attests to the unpredictability of predicting responsiveness to therapy by assaying for PITX2 methylation at *any* CpG dinucleotide. Specifically, Martens did not observe an association between PITX2 methylation in steroid receptor responsive tumors in patients who received tamoxifen as first-time treatment for recurrent breast cancer, and Nimmrich et al. states that in earlier work did not find PITX2 DNA methylation to be associated with intrinsic tamoxifen resistance in metastatic breast cancer. The Examiner states that Nimmrich studied DNA methylation of the PITX2 gene in untreated lymph node-negative hormone receptor positive breast cancer patients, finding that hypermethylation was associated with disease progression in these patients, and that methylation differences may occur between early and advanced breast cancer.

The Examiner states that the amount of experimentation required to practice the invention as claimed would be *undue* in view of the *Wands* factors.

Applicants' traversal:

Applicants respectfully traverse the Examiner's rejection in view of the following current responsive amendments to the claims:

Specifically, the claims have been limited to "human subject."

Additionally, the claims have been amended to recite "therapeutic treatment" in place of "therapy."

Moreover, independent claims 1, 45 and 62 have been amended to recite "wherein hypomethylation is indicative for a low risk for relapse while hypermethylation is indicative for a high risk for relapse, wherein predicting responsiveness of the subject to the therapeutic treatment is afforded." Support for the amendment is found throughout the originally filed specification, and particularly in Figure 19, in combination with the Table on page 58. Specifically, Figure 19 shows Kaplan-Meier curves of estimated disease free survival wherein the population of analyzed patients was split into two equal sized groups by their PITX2 methylation (p59 last paragraph). The survival of patients over time is displayed. The upper (dotted) line refers to patients which show hypomethylation of PITX2 (indicated by minus while the lower (continuous) line refers to patients which show hypermethylation of PITX2 (indicated by plus). Thus, more patients with hypo ethylated PITX2 are alive at a certain time point than patients with hyper ethylated PITX2. In other words, patients having hypo ethylated PITX2 have a lower risk for relapse (i.e. responders) than patients having hyper ethylated PITX2 (high risk of relapse i.e. non-responders). This is also evident for a person skilled in the relevant art in view of the section "Data set 2: Adjuvant setting" of Example 1, wherein the results of the Cox model are specified. As will be immediately evident to one of skill in the art, the positive value of the specified coefficient (coef) of the Cox model for PITX2 indicates that an increased risk for relapse correlates with hypermethylation of PITX2. In this regard, the description of Figure 19 on page 49 contains an inadvertent mistake, where, correctly, the upper curve shows responders and not non-responders, while the lower curve shows non-responders and not responders. This inadvertent mistake is obvious, because the corresponding patients of the upper curve lived longer than the patient of the

lower curve. The brief description of Figure 19 has been amended as described herein above to correct this inadvertent, obvious mistake.

With regard to the permissible scope of the breast tissue proliferative disorder genus, it is well recognized in the art that (1) a subclassification of breast cancer tissue into breast tissue subtypes is irrelevant for clinical implications (see attached literature citations to Sorlie et al., *PNAS*, 98:10869-10874, 2001, and Perou et al., *Nature* 406:747-752, 2000); and (2) all cells of the breast tissue originate from a common precursor. Moreover independent claims 1, 45 and 62 are already limited to a subclass of breast cancer namely estrogen receptor positive cells.

With regard to Martens et al. and Nimmrich et al. that Examiner has not presented any reason why these papers should preclude patentability of the presently amended claims.

Finally, claims 1, 45, 59, and 62 have been respectively limited to "SEQ ID NOS:83, 411, 412, 685, 686, complementary sequences or contiguous portions thereof." Complementarity" refers to complete complementarity as it is well known in the art. Applicants respectfully traverse the Examiner's position regarding commensurate claim scope with respect to the PITX2 gene limitation, and regarding alleged *undue* experimentation. The basis of Applicants' traversal in both instances fundamentally flows from the nature of the invention.

Specifically, in this case, the nature of the invention is premised on determination of CpG methylation within the PITX2 gene or its regulatory elements. In this regard, Applicants respectfully point out two art-recognized aspects of genomic methylation; namely (i) high-throughput methylation analysis, and (ii) the phenomenon of coordinate methylation (co-methylation) within genes and regulatory sequences.

High-throughput methylation analyses. By the priority date of the instant application, highly industrial, sophisticated array-based methylation assay procedures were available in the art that allow for the simultaneous analysis of the methylation status of thousands of CpG sequences for multiple indications in an efficient, high-throughput manner in a matter of days. Additionally, efficient high-throughput methods involving direct sequencing of bisulfite PCR products were known wherein the methylation present at any given CpG site is estimated by taking an average of

all fragments (thousands) generated during PCR resulting in statistically robust representation of the methylation present compared, for example, with subcloning (for example, this method was used in Eckhardt et al. (*Nat Genet.* 38(12):1378-85, 2006; Epub 2006 Oct 29); attached hereto, wherein CpG methylation over entire human chromosomes were efficiently analyzed. Significantly, as widely recognized in the art, these methods are not only efficient and high-throughput, but they are also routine—such that the nature of modern CpG methylation analysis is one of efficiency, wherein CpG methylation status over thousands of bases pairs can be routinely accomplished without undue experimentation in a matter of days or perhaps a few weeks.

Coordinate methylation (co-methylation). As appreciated in the art, CpG rich regions are commonly found in genes, and including in the regulatory sequences thereof. Moreover, as also appreciated in the art, for CpG methylation patterns within CpG rich regions (*e.g.*, such as those comprising the present PITX2 gene and/or its regulatory elements) methylation is typically found to be either present for all methylatable cytosines or none; in other words, increased/ hypermethylation of CpG dinucleotide sequences within CpG rich regions is typically *coordinate*, such that the hypermethylation status of a particular CpG within a CpG rich region can, with reasonable expectation, be extended to the other CpGs therein. For instance, Eckhardt et al. (*Nat Genet.* 38(12):1378-85, 2006; Epub 2006 Oct 29; and attached hereto) analyzed methylation (using bisulfite sequencing) in CG rich regions across entire chromosomes to provide a methylation map of the human genome. To date, these data comprise methylation data of 3 complete human chromosomes (22, 20, and 6) for a variety of different tissues and cell types. Based on these data, for methylation patterns within CpG rich regions (*e.g.*, such as those comprising the present PITX2 gene and regulatory elements) methylation is typically found to be either present for all methylatable cytosines or none. This methylation characteristic or pattern is referred to in the art as "co-methylation" or "coordinate methylation." The findings of this paper support a "significant correlation" of comethylation over thousands of nucleotides from any particular determined CpG (see, *e.g.*, page 2, column 2, 1st full paragraph, of attached Eckhardt et al publication document). Further confirmation of such co-methylation is provided by Taylor et

al. (*Cancer Res* 67:8511-8518, 2007; attached hereto), who confirm a pattern of concomitant methylation in 25 gene-related CpG rich regions taken from 40 neoplastic tissue isolates.

Such co-methylation in the context of the PITX2 gene is encompassed by Applicants' original conception and specification which teaches detection of methylated CpG dinucleotides within the PITX2 gene, including its promoter and regulatory regions. Indeed, such co-methylation underlies the basis for the MethyLight (Eads et al., *Cancer Re*s. 59:2302-2306, 1999; see also U.S. Patents 6,331,393 and 7,112,404) and HeavyMethyl (MethyLight assay is combined with methylation specific *blocking* probes covering CpG positions between the amplification primers) embodiments employed in the specification, as well as the bases for other long-standing art-recognized common methods such as MSP (Methylation-specific PCR); Herman et al. *Proc. Natl. Acad. Sci. USA* 93:9821-9826, 1996: US Patent No. 5,786,146), all of which rely on such co-methylation because the primers and/or probes used in these assays each typically encompass multiple CpG sequences, and which has now been further confirmed, for example, as part of the Eckhardt et al. data.

Therefore, based on the availability of high-throughput methylation analyses, and the nature of co-methylation that occurs within the PITX2 gene and its regulatory sequences, Applicants contend that while Applicants' specification and working Examples do not explicitly include analysis of each and every CpG position within the PITX2 gene and/or its regulatory elements, Applicants' method claims are nonetheless entitled to a broad scope with respect to the PITX2 gene limitation. A contrary conclusion would not comport with U.S. patent law on enablement.

Specifically, to satisfy the enablement requirement of 35 U.S.C. §112, first paragraph, the specification must teach one of skill in the art to make and use the invention without *undue* experimentation (*Atlas Powder Co.*), where this requirement can be satisfied by providing sufficient disclosure, either through illustrative examples or terminology, to teach one of skill in the art how to make and how to use the claimed subject matter without undue experimentation. This clause does <u>not</u> require "a specific example of everything within the scope of a broad claim"

(*In re Anderson*). A patentee is not only entitled to narrow claims particularly directed to the preferred embodiment, but also to broad claims which define the invention without a reference to specific instrumentalities (*Id*). Further, because "it is manifestly impracticable for an applicant who discloses a generic invention to give an example of every species falling within it, or even to name every such species, it is sufficient if the disclosure teaches those skilled in the art what the invention is and how to practice it" (*In re Grimme*). There is, therefore, no requirement for disclosure of every species within a genus. Applicants are entitled to claims that are commensurate in scope not only with what Applicants have specifically exemplified, but commensurate in scope with that which one of skill in the art could obtain by virtue of that which the Applicants have disclosed.

Applicants point out that under U.S. patent law, a considerable amount of experimentation is permissible, particularly if it is routine experimentation. As appreciated by the Examiner, the amount of experimentation that is permissible depends upon a number of factors (A, claim scope; B, nature of invention; C, state of the prior art; D, level of skill in the art; E, level of predictability; F, amount of direction provided; G, working examples; and H, quantity of experimentation required) as discussed herein above. The Examiner has offered insufficient evidence to support that any alleged amount of experimentation is other than rapid, high-throughput and routine.

With respect to these factors in the present case, the Examiner appreciates that the level of skill in the art is very high (as evidenced by the Examiner's own cited art references), and given the nature of the invention in terms of the realities of high-throughput methylation assays and co-methylation within the PITX2 gene and its regulatory sequences, Applicants contend that a *prima facie* case of insufficient enablement cannot reasonably be supported under U.S. Patent law, because under the proper analysis of *all* Wands factors, any amount of experimentation required to practice the invention as presently claimed would in fact be merely routine, and insufficient to support the Examiner's allegation of *undue* experimentation. Improper limitation of Applicants' invention to particular exemplary preferred regions within the PITX2 gene is not only impermissible under U.S. patent law in view of the present facts, but would also be unjustifiable—

in that a person of ordinary skill in the art could, using routine, efficient methods readily identify and select alternate diagnostic CpG positions within the PITX2 gene and its regulatory sequences but outside Applicants' exemplary preferred regions, thus effectively eviscerating Applicants' claimed invention.

Applicants are entitled to claims that are commensurate in scope not only with what Applicants have specifically exemplified, but commensurate in scope with that which one of skill in the art could obtain by virtue of that which the Applicants have disclosed.

In view of the present claim amendments, Applicants respectfully contend that the currently amended claim scope is commensurate with the teachings of the specification, and request withdrawal of the Examiner's rejection based on lack of sufficient enablement.

Obviousness-type Double Patenting Rejection

The Examiner has *provisionally* rejected claims 1, 17-24, 45, 57-59, 61, 62, 67, and 77, on the grounds of nonstatutory obviousness-type double patenting, as being unpatentable over claims 1, 3-6, 8-11, and 20-21 of Applicants' copending Application No. 10/582,705.

Applicants respectfully traverse this rejection based on the fact that in view of the prior art (including that specifically cited and discussed by the Examiner in the present Office Action), no *prima facie* case of obviousness can be supported. Specifically, "complementarity" as recited in the present claims, refers to complete complementarity as it is well known in the art, such that the Examiner's reliance on "complementarity" to SEQ ID NO:23 of '705 is not reasonably supported.

Obviously, if required depending on the allowed claim language, Applicants are prepared to file a Terminal Disclaimer if necessary under the facts.

However, in view of the present facts, Applicants respectfully request withdrawal of this rejection.

CONCLUSION

In view of the foregoing amendments and remarks, Applicants respectfully request entry of the present Response and Amendment and allowance of all claims as provided herein above. The Examiner is encouraged to phone Applicants' attorney, Barry L. Davison, to resolve any outstanding issues and expedite allowance of this application.

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